**INTRODUCTION**

Automated liquid handling was optimized for improving LC-MS/MS analysis (1-3). Samples were digested with trypsin (1:200) overnight at 37 °C. For automatic sample processing, we developed a method for VIAFLO 96 from Integra and other high-throughput automatic liquid handling systems (Figure 2). Desalting peptides is an essential procedure for improving LC-MS/MS analysis (1-3). Removing salts utilizes solid phase extraction (SPE) techniques, but these procedures are not readily adapted for automated liquid handling systems. However, we have automated, disposable pipette tips containing IMCStips for efficient desalting operations.

**RESULTS**

Automatic sample preparation using VIAFLO 96 from Integra and a high-throughput automatic liquid handling system was optimized for VIAFLO 96 from Integra and other high-throughput automatic liquid handling systems (Figure 2). Desalting IMCStips with RP resin on a VIAFLO96 from Integra system was used for automatic sample preparation. For global peptide identification followed by desalting, we used Q-Exactive mass spectrometer coupled with Ultimate 3000 nano-UHPLC system. MS spectra were acquired by data dependent scans consisting of MS/MS scans of the twenty most abundant ions. To measure desalted peptide quantity, we used UPLC TSQ-Endura triple quadrupole mass spectrometry with optimized conditions.

**CONCLUSIONS**

Mass spectrometry has become a mainstream analytical tool for a broad range of applications. One of the major bottlenecks in mass spectrometry is the ability to process many samples in a consistent and reproducible manner. This consistency should leverage an automated liquid handling system that eliminates many of the errors from non-automated manual operations. Here, we explored several different resin types with discrete dried extraction tips packaging (IMCStips) on an automated liquid handler. This approach demonstrates faster worktimes while exhibiting higher recovery efficiencies than traditional spin column formats. Furthermore, the screening of several different resins (C18, silica, graphitic carbon black and two polystyrene) was done to determine the most effective resin for routine desalting and peptide enrichment. Based on the work, IMCStips packed with 50 μm polystyrene crosslinked with divinylbenzene (named as RP) showed consistently high recoveries of the control peptides, phosphopeptides and higher protein IDs from cell lysates. The peptide recovered using the RP resin showed little or no statistical variation from the peptide recovered using C18 resin as indicated by GRAVY value. The flexibility and high throughput capabilities of IMCStips for peptide processing applications were relative to the successful large number of samples while maintaining highly consistent operations.

**REFERENCES**


Abbreviations:

ACN: Acetonitrile; AUC: Area under the curve; C18-C4: C18-C4 tips; C18-C4: C18-C4 spin column from Thermo Fisher; RP: polystyrene/divinylbenzene crosslinked with divinylbenzene; FFA: Free fatty acid; GC/MS: Gas Chromatography/Mass Spectrometry.